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Wounds

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14. ABSTRACT

The complexity and severity of the extremity wounds seen in recent military operations have necessitated the demand for improved wound assessment, before, during and after medical treatment. We have quantified parameters such as tissue oxygenation, tissue temperature and the molecular environment at specific locations within the wound, to better understand the wound healing process in various wound phenotypes. This has helped us to establish which parameters are most important for a correct initial assessment of the wound along with a more accurate wound healing prediction. We will apply the same research strategy to wound healing in a civilian trauma center. The parameters bearing the most weight will be used to diagnose trauma wounds and predict patient outcome.

15. SUBJECT TERMS
 Wound healing; biomarkers; Raman spectroscopy; 3CCD imaging; infrared imaging; tissue oxygenation

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INTRODUCTION

Casualties in Operation Iraqi Freedom (OIF) and Operation Enduring Freedom (OEF) have experienced a high rate of extremity injuries with nearly ubiquitous diffuse tissue damage and compromised local circulation often associated with overt vascular injury. These injuries include traumatic amputations, open fractures, crush injuries, burns, acute vascular disruption, blastwave-associated pressure injuries, air, thrombotic, and fat embolism, and compartment syndrome. In the treatment of such complex traumatic injuries, improved assessment of global and regional perfusion, extent of infection, location and development of necrotic tissue, as well as location and development of early heterotopic ossification would facilitate the resuscitation and definitive treatment of these patients. Noninvasive spectroscopic methods may fulfill such a role, particularly Raman spectroscopy, infrared imaging, and visible reflectance spectroscopic imaging. These technologies are capable of monitoring tissue temperature (1), perfusion (2) and associated hypoxia (3-6), collagen deposition (7, 8), and development of calcified tissue (9-18).

The complexity and severity of the extremity wounds seen in recent military operations have necessitated the demand for improved wound assessment, before, during and after medical treatment. We have quantified parameters such as tissue oxygenation, tissue temperature and the molecular environment at specific locations within the wound, to better understand the wound healing process in various wound phenotypes. This has helped us to establish which parameters are most important for a correct initial assessment of the wound along with a more accurate wound healing prediction. The parameters bearing the most weight will be used to diagnose trauma wounds and predict patient outcome. Improved objective assessment of acute wounds would be conducive to improved treatment of the wounds which may result in faster healing times, decreased infection rates, and decreased local and systemic complications of injury.

KEYWORDS

Wound healing; biomarkers; Raman spectroscopy; 3CCD imaging; infrared imaging; tissue oxygenation

OVERALL PROJECT SUMMARY

Our hypothesis is that the development of an integrated prediction model (using spectroscopy and spectroscopic imaging data in addition to clinical data) will allow for more accurate assessment of tissue perfusion and oxygenation in extremity injuries, providing improved diagnosis and prognosis of the affected tissue. Improved objective assessment of acute wounds would be conducive to improved treatment of the wounds which may result in faster healing times, decreased infection rates, and decreased local and systemic complications of injury.

This proposal will serve to validate these prediction models, developed from a military population of acute combat wounds, in civilian acute wounds. We have initiated a collaboration with Emory University to expand this study at a civilian Level 1 trauma center.

Aim 1
Aim 1 is comprised of four tasks:

a) Collect 3-CCD and IR images of extremity wounds in the operating room.

b) Collect serum, wound exudates, and tissue biopsies from each wound during the surgical debridement process.

c) Perform quantitative bacteriology on wound effluent and tissue, RT-PCR on tissue biopsies, protein analysis on serum, and Raman spectroscopy on tissue biopsies and wound effluent.

d) Correlate the presence of necrotic tissue (depth and breadth), wound infection (serial quantitative bacteriology, development of heterotopic ossification, physician and pathologist observations, and serum cytokine levels and wound cytokine and gene expression levels. (n=100)

To date, we have enrolled 18 patients at Grady Memorial Hospital through Emory University. Only one eligible patient has declined enrollment. Initial enrollment was extremely slow and limited our ability to complete Tasks 1a-d; as of February 2014 we had only enrolled 8 patients into the study. Slow enrollment was due to inclusion criteria not being met by patients that were not vulnerable persons, and an inability to enroll patients that did not speak English. Since amending the clinical protocol to include a consent form in Spanish, we have been able to enroll an additional 10 patients in only 3 months. We anticipate that enrollment numbers will continue to grow, particularly through the summer months.

To support the execution of Task 1a, we have provided the clinical team at Emory University with an additional 3CCD camera in anticipation of increased enrollment rates.

As outlined in Task 1b, serum, wound exudate, and tissue biopsies have been collected for each patient enrolled in the study to date. As samples are received from Emory

University and processed, we are collecting quantitative bacteriology on wound effluent and tissue, RT-PCR on tissue biopsies, and protein analysis on serum (Task 1c).

Though task 1c also specifies the collection of Raman spectra of tissue biopsies and wound effluent, the 830 nm Raman spectroscopic system necessary to collect the spectra was not installed until April 2014. Since the instrument's installation, however, Raman spectra are being collected (5 replicates per biopsy and effluent sample). When comparing Raman spectra of the same sample collected on the 785 nm and 830 nm Raman spectroscopic systems, the fluorescence seen in the spectrum collected on the 785 nm system (Figure 1A) is significantly reduced in the Raman spectrum collected on the new 830 nm system (Figure 1B). This inherently increases the signal-to-noise ratio of the 830 nm Raman spectrum, enabling collection of a superior Raman spectrum.

Task 1d cannot be completed until Task 1a-c are completed. We will conduct a preliminary analysis of the meta-data when enrollment reaches 25 patients at Emory University.

Aim 2
Aim 2 is revolves around the use machine learning to validate spectroscopic and clinical results with patient outcome as a predictor. In this aim, we will predict patient outcomes based on correlations between spectroscopic and clinical parameters and wound grade and healing. (n=100 patients). We are currently still enrolling patients and have not yet amassed sufficient data to begin evaluating the use of machine learning to validate spectroscopic and clinical results with patient outcome as a predictor. As with Task 1d, we will perform an initial evaluation of machine learning prediction models when enrollment reaches 25 patients.

KEY RESEARCH ACCOMPLISHMENTS

- We have received serum, wound exudate, and tissue biopsies from 18 patients to date.

- The new 830 nm Raman spectroscopic system has been installed and verified. Fluorescence background that previously overwhelmed some of our Raman spectra has been significantly reduced, enabling the collection of Raman spectra with superior signal-to-noise ratios.

- Efforts to collect Raman spectra of colonized wound effluent samples advances.

- We have applied for and been awarded a no cost extension of the award until May 2015. This should enable our projected patient enrollment for this study (n=100).

CONCLUSION

This project was significantly delayed due to slow enrollment and late receipt of the new 830 nm Raman spectroscopic system. Both of these issues have since been mitigated. We have since enrolled 18 patients and continue to move forward the research. Having received a no cost extension of the award, anticipate meeting our originally stated goal of addressing key decision points for critically ill trauma and surgical patients including: 1) timing of wound closure; 2) assessment of bacterial bioburden, antibiotic utilization and treatment response, and subsequent infections; 3) techniques of wound coverage and management; management of segmental bone defects; 4) amputee-specific issues including, timing of wound closure, management of infection, ischemia and heterotopic ossification; 5) heterotopic ossification mechanisms, characteristics, risk stratification, treatment, and prevention; and 6) risk stratification, prevention, and management of venous thromboembolism (VTE).

PUBLICATIONS, ABSTRACTS, AND PRESENTATIONS:

Lay Press
NOTHING TO REPORT.

Peer-Reviewed Scientific Journals
NOTHING TO REPORT.

Invited Articles
NOTHING TO REPORT.

Abstracts
NOTHING TO REPORT.

Presentations
NOTHING TO REPORT.

INVENTIONS, PATENTS AND LICENSES

NOTHING TO REPORT.

REPORTABLE OUTCOMES

Nothing to report.

OTHER ACHIEVEMENTS

Nothing to report.

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APPENDICES
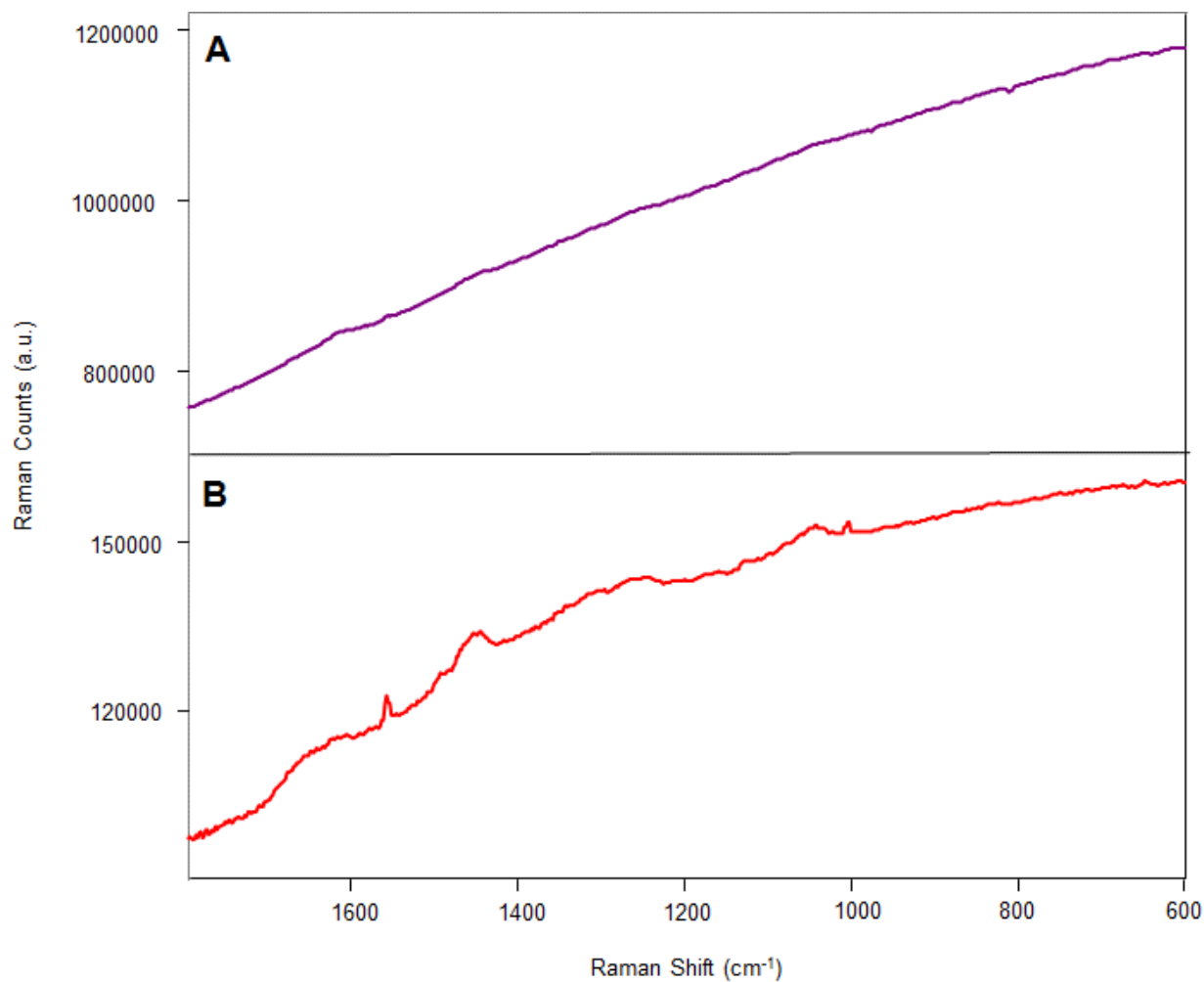
Figure 1 – Comparison of spectra collected on the 785 nm Raman spectroscopic system (A) and the new 830 nm Raman spectroscopic system (B).